

What is claimed is:

1. An array of protein-capture agents, comprising:
 - (a) a substrate;
 - (b) at least one organic thinfilm covering some or all of the surface of the substrate; and
 - (c) a plurality of patches arranged in discrete, known regions on the portions of the substrate surface covered by organic thinfilm, wherein:
 - (i) each patch comprises protein-capture agents immobilized on the organic thinfilm, wherein said protein-capture agents of a given patch are capable of binding a particular expression product, or a fragment thereof, of a cell or population of cells in an organism; and
 - (ii) said array comprises a plurality of different protein-capture agents, each of which is capable of binding a different expression product, or fragment thereof, of the cell or population of cells.
2. The array of Claim 1 which comprises at least about 10 of said patches.
3. The array of Claim 2 which comprises at least about 100 of said patches.
4. The array of Claim 3 which comprises at least about 10^3 of said patches.

5. The array of Claim 1 which comprises at least about 10 different protein-capture agents.
6. The array of Claim 5 which comprises at least about 100 different protein-capture agents.
7. The array of Claim 6 which comprises at least about 1000 different protein-capture agents.
8. The array of Claim 1, wherein the area of the substrate surface covered by each of the patches is no more than about 0.25 mm^2 .
9. The array of Claim 8, wherein the area of the substrate surface covered by each of the patches is between about $1 \text{ }\mu\text{m}^2$ and about $10,000 \text{ }\mu\text{m}^2$.
10. The array of Claim 1, wherein the patches are all contained within an area of about 1 cm^2 or less on the surface of the substrate.
11. The array of Claim 1, wherein the protein-capture agents are proteins.

12. The array of Claim 11, wherein the protein-capture agents are antibodies or antibody fragments.

13. The array of Claim 12, wherein the antibodies or antibody fragments have been derived by selection from a library using the phage display method.

14. The array of Claim 13, wherein the antibodies or antibody fragments have been derived by affinity binding to the proteins of a cellular extract or body fluid.

15. The array of Claim 12, wherein said antibodies or antibody fragments are selected from the group consisting of monoclonal antibodies, Fab fragments, and single-chain Fvs.

16. The array of Claim 1, wherein the organic thinfilm on the array is less than about 20 nm thick.

17. The array of Claim 1, wherein the organic thinfilm on the array comprises a monolayer.

18. The array of Claim 17, wherein the monolayer comprises a self-assembled monolayer comprising molecules of the formula



wherein R is a spacer, X is a functional group that binds R to the surface, Y is a functional group for binding the protein-capture agent onto the monolayer, and a and b are, independently, integers.

19. The array of Claim 18, wherein both a and b are equal to 1.

20. The array of Claim 18, wherein:

said substrate is selected from the group consisting of silicon, silicon dioxide, indium tin oxide, alumina, glass, and titania; and

X, prior to incorporation into said monolayer, is selected from the group consisting of a monohalosilane, dihalosilane, trihalosilane, trichlorosilane, trialkoxysilane, dialkoxysilane, monoalkoxysilane, carboxylic acid, and phosphate.

21. The array of Claim 18, wherein the substrate comprises silicon and X is an olefin.

22. The array of Claim 1, wherein the substrate comprises a polymer.

23. The array of Claim 18, further comprising at least one coating between the substrate and the monolayer, wherein said coating is formed on the substrate or applied to the substrate.

24. The array of Claim 23, wherein:

the coating is a noble metal film; and

X, prior to incorporation into said monolayer, is a functional group selected from the group consisting of an asymmetrical or symmetrical disulfide, sulfide, diselenide, selenide, thiol, isonitrile, selenol, trivalent phosphorus compounds, isothiocyanate, isocyanate, xanthanate, thiocarbamate, phosphines, amines, thio acid and dithio acid.

25. The array of Claim 23, wherein the coating is titania or tantalum oxide and X is a phosphate group.

26. The array of Claim 1, wherein each protein-capture agent has been immobilized onto the organic thinfilm by an affinity tag.

27. An array of bound proteins, comprising:

(a) the array of Claim 1; and

(b) a plurality of different proteins which are expression products, or fragments thereof, of a cell or a population of cells in an organism, wherein each of said different proteins is bound to a protein-capture agent on a separate patch of the array.

28. A diagnostic device comprising the array of Claim 1.

29. A method of assaying in parallel for a plurality of different proteins in a sample which are expression products, or fragments thereof, of a cell or a population of cells in an organism, comprising:

(a) delivering the sample to an array of spatially distinct patches of different protein-capture agents under conditions suitable for protein binding, wherein each of the proteins being assayed is a binding partner of the protein-capture agent of at least one patch on the array; and

(b) detecting, either directly or indirectly, for the presence or amount of protein bound to each patch of the array.

30. A method of assaying in parallel for a plurality of different proteins in a sample which are expression products, or fragments thereof, of a cell or a population of cells in an organism, comprising:

(a) delivering the sample to an array of Claim 1 under conditions suitable for protein binding, wherein each of the proteins being assayed is a binding partner of the protein-capture agent of at least one patch on the array; and

(b) detecting, either directly or indirectly, for the presence or amount of protein bound to each patch of the array.

31. The method of claim 30, further comprising the step:

further characterizing the proteins bound to at least one patch of the array.

32. The method of Claim 31, wherein said step of further characterizing the proteins comprises measuring the activity of the proteins.

33. A method for determining the protein expression pattern of a cell or a population of cells in an organism, comprising:

(a) delivering a sample containing the expression products, or fragments thereof, of the cell or population of cells to an array of Claim 1 under conditions suitable for protein binding, and

(b) detecting, either directly or indirectly, for the amount of protein bound to each patch of the array.

34. A method of comparing the protein expression patterns of two cells or populations of cells, comprising:

(a) delivering a sample containing the expression products, or fragments thereof, of a first cell or population of cells to a first array of Claim 1 under conditions suitable for protein binding;

(b) delivering a sample containing the expression products, or fragments thereof, of a second cell or population of cells to a second array, wherein the second array is identical to the first array;

(c) detecting, either directly or indirectly, for the amount of protein bound to each patch on the washed first and second arrays; and

(d) comparing the amounts of protein bound to the patches of the first array to the amounts of protein bound to the corresponding patches of the second array.

35. A method of evaluating a disease condition in a tissue in an organism, comprising:

(a) contacting a sample comprising the expression products, or fragments thereof, of the cells of the tissue being evaluated with an array of Claim 1 under conditions suitable for protein binding, wherein the binding partners of a plurality of protein-capture agents on the array include proteins which are expression products, or fragments thereof, of the cells of the tissue and whose expression levels are indicative of the disease condition; and

(b) detecting, directly or indirectly, for the amount of protein bound to each patch of the array.

36. A method for producing the array of Claim 1, comprising:

(a) selecting recombinant bacteriophage expressing antibody fragments from a phage display library, wherein said recombinant bacteriophage are selected by affinity binding to a protein which is an expression product, or fragment thereof, of a cell or population of cells in an organism;

(b) producing at least one purified sample of an antibody fragment from a bacteriophage selected in step (a); and

(c) repeating steps (a)-(b) with a different proteins which are expression products, or fragments thereof, of a cell or population of cells from the organism, or a fragment of the second protein, until the desired plurality of purified samples of different antibody fragments with different binding pairs is produced; and

(d) immobilizing the antibody fragment of each different purified sample onto an organic thinfilm on a separate patch on the surface of a substrate to form a plurality of patches of antibody fragments on discrete, known regions of the substrate surface.

37. A method for producing an array of protein-capture agents, comprising:

(a) selecting protein-capture agents from a library of protein-capture agents, wherein the protein-capture agents are selected by their binding affinity to the proteins in a cellular extract or body fluid;

(b) producing a plurality of purified samples of the selected protein-capture agents of step (a); and

(c) immobilizing the protein-capture agent of each different purified sample onto an organic thinfilm on a separate patch on the surface of a substrate to form a plurality of patches of protein-capture agents on discrete, known regions of the substrate surface.

38. The method of Claim 37, wherein said protein-capture agents are antibody fragments displayed on the surface of recombinant bacteriophages and said library of protein-capture agents is a phage display library.

39. A method of Claim 38, further comprising:

biasing the library of protein-capture agents by eliminating from the library those protein-capture agents which bind certain proteins, wherein the protein-capture agents which are eliminated are removed from the library by their binding affinity to said certain proteins.

40. The method of claim 39, wherein said certain proteins are proteins in a second cellular extract or body fluid.

41. An array of protein-capture agents produced by the method of Claim 37.

42. A method for producing an array of protein-capture agents, comprising:

(a) selecting protein-capture agents from a library of protein-capture agents, wherein the protein-capture agents are selected by their binding affinity to proteins which are the expression products, or fragments thereof, of a cDNA expression library;

(b) producing a plurality of purified samples of the protein-capture agents of step (a); and

(c) immobilizing the protein-capture agent of each different purified sample onto an organic thinfilm on a separate patch on the surface of a substrate to form a plurality of patches of protein-capture agents on discrete, known regions of the surface of the substrate.

43. The method of Claim 42, wherein said protein-capture agents are antibody fragments displayed on the surface of recombinant bacteriophages and said library of protein-capture agents is a phage display library.

44. A method of Claim 42, further comprising:

 biasing the library of protein-capture agents by eliminating from the library those protein-capture agents which bind certain proteins, wherein the protein-capture agents which are eliminated are removed from the library by their binding affinity to said certain proteins.

45. An array of protein-capture agents produced by the method of Claim 42.